Otor: CANIGGIA ET AL. GETHODS TO DIAGNOSE A REQUIRED REGULATION OF T No.: 11757.38USD1 BLAST Serial No.: 10/028,158 Sheet 1 of 21

#### FIG. 1

```
12-SEP-1993
                                         RNA
                Human mRNA for transforming growth factor-beta 3 (TGF-beta
                             2574 bp
                HSTGFB3M
LOCUS
DEFINITION
                3).
                X14149
ACCESSION
                growth factor; transforming growth factor; transforming
NID
KEYWORDS
                growth factor-beta 3.
                human.
SOURCE
                Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
  ORGANISM
                 Vertebrata; Eutheria; Primates; Catarrhini; Hominidae;
                 Homo.
                 1 (bases 1 to 2574)
REFERENCE
                 Chen. E.Y.
   AUTHORS
                 Direct Submission
                 Submitted (23-MAR-1989) Chen E.Y., Genentech Inc., 460 Pt.
   TITLE
                 San Bruno Blvd., San Francisco, CA 94080, USA
   JOURNAL
                 2 (bases 1 to 2574)
                 Derynck, R., Lindquist, P.B., Lee, A., Wen, D., Tamm, J.,
 REFERENCE
                 Graycar, J.L., Rhee, L., Mason, A.J., Miller, D.A.,
   AUTHORS
                 Coffey, R.J., Moses, H.L. and Chen, E.Y.
                 A new type of transforming growth factor-beta, TGF-beta 3
   TITLE
                 EMBO J. 7 (12), 3737-3743 (1988)
   JOURNAL
                  89091120
                  See <J03241> for alternative sequence of TGF-beta 3.
   MEDLINE
 COMMENT
                        Location/Qualifiers
  FEATURES
                        1..2574
       source
                        /organism="Homo sapiens"
                              /db_xref="taxon:9606"
                               /tissue_type=*placenta, ovary glioblastoma*
                               /cell_line="A172 glioblastoma"
                               /chromosome="14q24"
                        254..1492
       CDS
                               /note="TGF-beta 3 (AA 1-412)"
                               /codon_start=1
                               /db_xref="PID:g37096"
                               /db_xref="SWISS-PROT:P10600"
                   /translation="MKMHLQRALVVLALLNFATVSLSLSTCTTLDFGHIKKKRVEAIR
                   GQILSKLRLTSPPEPTVMTHVPYQVLALYNSTRELLEEMHGEREEGCTQENTESEYYA
                   KEIHKFDMIQGLAEHNELAVCPKGITSKVFRFNVSSVEKNRTNLFRAEFRVLRVPNPS
                   {\tt SKRNEQRIELFQILRPDEHIAKQRYIGGKNLPTRGTAEWLSFDVTDTVREWLLRRESN}
                   LGLEISIHCPCHTFQPNGDILENIHEVMEIKFKGVDNEDDHGRGDLGRLKKQKDHHNP
                   \verb|HLILMMIPPHRLDNPGQGGQRKKRALDTNYCFRNLEENCCVRPLYIDFRQDLGWKWVH\\
                   \tt EPKGYYANFCSGPCPYLRSADTTHSTVLGLYNTLNPEASASPCCVPQDLEPLTILYYV
                   GRTPKVEQLSNMVVKSCKCS"
                                                599 t
                                       666 g
                             680 c
```

629 a

BASE COUNT

11

Inventor: CANIGGIA ET AL.
pcket No.: 11757.38USD1
tle: METHODS TO DIAGNOSE A REQUIRED REGULATION (
PHOBLAST
INVASION
Serial No.: 10/028,158
Sheet 2 of 21

#### FIG. 1 (cont'd)

```
ORTGIN
    1 cctgtttaga cacatggaca acaatcccag cgctacaagg cacacagtcc gcttcttcgt
   61 cctcagggtt gccagcgctt cctggaagtc ctgaagctct cgcagtgcag tgagttcatg
  121 caccttcttg ccaagcctca gtctttggga tctggggagg ccgcctggtt ttcctccctc
  181 cttctgcacg tctgctgggg tctcttcctc tccaggcctt gccgtccccc tggcctctct
  241 teccagetea cacatgaaga tgeaettgea aagggetetg gtggteetgg eeetgetgaa
  301 ctttgccacg gtcagcctct ctctgtccac ttgcaccacc ttggacttcg gccacatcaa
  361 gaagaagagg gtggaagcca ttaggggaca gatcttgagc aagctcaggc tcaccagccc
  421 ccctgagcca acggtgatga cccacgtccc ctatcaggtc ctggcccttt acaacagcac
  481 ccgggagctg ctggaggaga tgcatgggga gagggaggaa ggctgcaccc aggaaaacac
  541 cgagtcggaa tactatgcca aagaaatcca taaattcgac atgatccagg ggctggcgga
  601 gcacaacgaa ctggctgtct gccctaaagg aattacctcc aaggttttcc gcttcaatgt
  661 gtcctcagtg gagaaaaata gaaccaacct attccgagca gaattccggg tcttgcgggt
  721 geccaacce agetetaage ggaatgagea gaggategag etettecaga teetteggee
  781 agatgagcac attgccaaac agcgctatat cggtggcaag aatctgccca cacggggcac
  841 tgccgagtgg ctgtcctttg atgtcactga cactgtgcgt gagtggctgt tgagaagaga
  901 gtccaactta ggtctagaaa tcagcattca ctgtccatgt cacacctttc agcccaatgg
  961 agatatcctg gaaaacattc acgaggtgat ggaaatcaaa ttcaaaggcg tggacaatga
 1021 ggatgaccat ggccgtggag atctggggcg cctcaagaag cagaaggatc accacaaccc
 1081 tcatctaatc ctcatgatga ttcccccaca ccggctcgac aacccgggcc aggggggtca
 1141 gaggaagaag cgggctttgg acaccaatta ctgcttccgc aacttggagg agaactgctg
 1201 tgtgcgcccc ctctacattg acttccgaca ggatctgggc tggaagtggg tccatgaacc
 1261 taagggctac tatgccaact tctgctcagg cccttgccca tacctccgca gtgcagacac
 1321 aacccacage acggtgctgg gactgtacaa cactctgaac cctgaagcat ctgcctcgcc
 1381 ttgctgcgtg ccccaggacc tggagcccct gaccatcctg tactatgttg ggaggacccc
 1441 caaagtggag cagctctcca acatggtggt gaagtcttgt aaatgtagct gagaccccac
 1501 gtgcgacaga gagaggggag agagaaccac cactgcctga ctgcccgctc ctcgggaaac
 1561 acacaagcaa caaacctcac tgagaggcct ggagcccaca accttcggct ccgggcaaat
 1621 ggctgagatg gaggtttcct tttggaacat ttctttcttg ctggctctga gaatcacggt
 1681 ggtaaagaaa gtgtgggttt ggttagagga aggctgaact cttcagaaca cacagacttt
 1741 ctgtgacgca gacagagggg atggggatag aggaaaggga tggtaagttg agatgttgtg
 1801 tggcaatggg atttgggcta ccctaaaggg agaaggaagg gcagagaatg gctgggtcag
 1861 ggccagactg gaagacactt cagatetgag gttggatttg etcattgetg taccacatet
 1921 gctctaggga atctggatta tgttatacaa ggcaagcatt tttttttta aagacaggtt
 1981 acgaagacaa agtoccagaa tigtatotoa tactgtotgg gattaagggo aaatotatta
 2041 cttttgcaaa ctgtcctcta catcaattaa catcgtgggt cactacaggg agaaaatcca
 2101 ggtcatgcag ttcctggccc atcaactgta ttgggccttt tggatatgct gaacgcagaa
 2161 gaaagggtgg aaatcaaccc tctcctgtct gccctctggg tccctcctct cacctctccc
 2221 togatcatat ttccccttgg acacttggtt agacgccttc caggtcagga tgcacatttc
 2281 tggattgtgg ttccatgcag ccttggggca ttatgggtct tcccccactt cccctccaag
 2341 accetgtgtt catttggtgt teetggaage aggtgetaca acatgtgagg cattegggga
 2401 agctgcacat gtgccacaca gtgacttggc cccagacgca tagactgagg tataaagaca
 2461 agtatgaata ttactctcaa aatctttgta taaataaata tttttggggc atcctggatg
 2521 atttcatctt ctggaatatt gtttctagaa cagtaaaagc cttattctaa ggtg
```

Inventor: CANIGGIA ET AL.

cket No.: 11757.38USD1

EMETHODS TO DIAGNOSE A REQUIRED REGULATION OF HOBLAST

/ASION

Serial No.: 10/028.158

Sheet 3 of 21

#### FIG. 2

```
LOCUS
             HSU22431
                          3678 bp
                                     mRNA
                                                      PRI
                                                                 28-JUN-1995
             Human hypoxia-inducible factor 1 alpha (HIF-1 alpha) mRNA, complete
 DEFINITION
             cds.
ACCESSION
             U22431
NID
             g881345
KEYWORDS
SOURCE
             human.
  ORGANISM
             Homo sapiens
             Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
             Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE
                (bases 1 to 3678)
  AUTHORS
             Wang, G.L., Jiang, B.H., Rue, E.A. and Semenza, G.L.
  TITLE
             Hypoxia-inducible factor 1 is a basic-helix-loop-helix-PAS
             heterodimer regulated by cellular O2 tension
  JOURNAL
             Proc. Natl. Acad. Sci. U.S.A. 92 (12), 5510-5514 (1995)
  MEDLINE
             95296340
REFERENCE
                (bases 1 to 3678)
  AUTHORS
             Wang, G.L., Jiang, B.-H., Rue, E.A. and Semenza, G.L.
  TITLE
             Direct Submission
             Submitted (09-MAR-1995) Gregg L. Semenza, Center for Medical
  JOURNAL
            Genetics, The Johns Hopkins University School of Medicine, 600 N.
            Wolfe St., Baltimore, MD 21287-3914, USA
FEATURES
                      Location/Qualifiers
     source
                      1..3678
                      /organism="Homo sapiens"
                      /db xref="taxon:9606"
                      /cell line="Hep3B"
                      /cell_type="hepatoblastoma"
                      29..2509
     gene
                      /gene="HIF-1 alpha"
     CDS
                      29..2509
                      /gene="HIF-1 alpha"
                      /standard_name="hypoxia-inducible factor 1, alpha subunit"
                      /note="basic helix-loop-helix transcription factor"
                      /codon_start=1
                      /product="hypoxia-inducible factor l alpha"
                      /db xref="PID:g881346"
                     /translation="MEGAGGANDKKKISSERRKEKSRDAARSRRSKESEVFYELAHQL
                     PLPHNVSSHLDKASVMRLTISYLRVRKLLDAGDLDIEDDMKAQMNCFYLKALDGFVMV
                     LTDDGDMIYISDNVNKYMGLTQFELTGHSVFDFTHPCDHEEMREMLTHRNGLVKKGKE
                     QNTQRSFFLRMKCTLTSRGRTMNIKSATWKVLHCTGHIHVYDTNSNQPQCGYKKPPMT
                     CLVLICEPIPHPSNIEIPLDSKTFLSRHSLDMKFSYCDERITELMGYEPEELLGRSIY
                     EYYHALDSDHLTKTHHDMFTKGQVTTGQYRMLAKRGGYVWVETQATVIYNTKNSQPQC
                     IVCVNYVVSGIIQHDLIFSLQQTECVLKPVESSDMKMTQLFTKVESEDTSSLFDKLKK
                     EPDALTLLAPAAGDTIISLDFGSNDTETDDQQLEEVPLYNDVMLPSPNEKLQNINLAM
                     SPLPTAETPKPLRSSADPALNQEVALKLEPNPESLELSFTMPQIQDQTPSPSDGSTRQ
                     SSPEPNSPSEYCFYVDSDMVNEFKLELVEKLFAEDTEAKNPFSTQDTDLDLEMLAPYI
                     PMDDDFQLRSFDQLSPLESSSASPESASPQSTVTVFQQTQIQEPTANATTTTATTDEL
                     KTVTKDRMEDIKILIASPSPTHIHKETTSATSSPYRDTQSRTASPNRAGKGVIEQTEK
                     SHPRSPNVLSVALSQRTTVPEEELNPKILALQNAQRKRKMEHDGSLFQAVGIGTLLQQ
                     PDDHAATTSLSWKRVKGCKSSEQNGMEQKTIILIPSDLACRLLGQSMDESGLPQLTSY
                     DCEVNAPIQGSRNLLQGEELLRALDQVN*
     polyA_site
                     3678
                     /note="42 A nucleotides"
BASE COUNT
               1197 a
                          695 c
                                   675 g
```

Inventor: CANIGGIA ET AL.
por et No.: 11757.38USD1
METHODS TO DIAGNOSE A REQUIRED REGULATION OF TISION
Serial No.: 10/028,158
Sheet 4 of 21

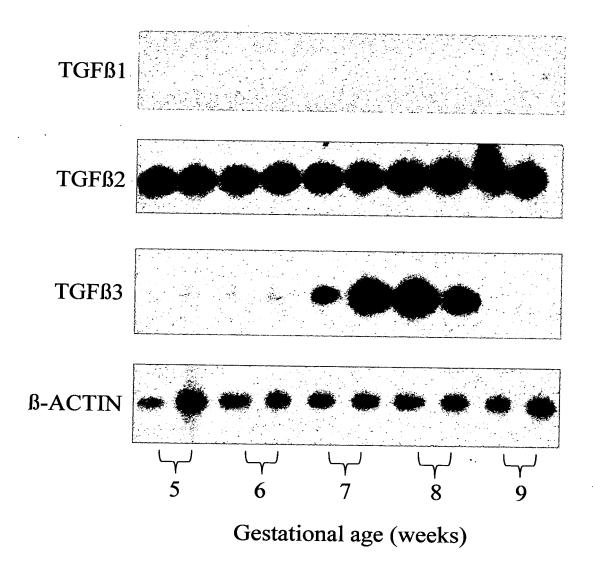
#### FIG. 2 (cont'd)

#### ORIGIN

```
1 gtgaagacat cgcggggacc gattcaccat ggagggcgcc ggcggcgcga acgacaagaa
  61 aaagataagt tetgaaegte gaaaagaaaa gtetegagat geageeagat eteggegaag
 121 taaagaatct gaagtttttt atgagettge teateagttg ceaetteeae ataatgtgag
 181 ttcgcatctt gataaggcct ctgtgatgag gcttaccatc agctatttgc gtgtgaggaa
 241 acttctggat gctggtgatt tggatattga agatgacatg aaagcacaga tgaattgctt
 301 ttatttgaaa goottggatg gttttgttat ggttotcaca gatgatggtg acatgattta
 361 catttctgat aatgtgaaca aatacatggg attaactcag tttgaactaa ctggacacag
 421 tgtgtttgat tttactcatc catgtgacca tgaggaaatg agagaaatgc ttacacacag
 481 aaatggcctt gtgaaaaagg gtaaagaaca aaacacacag cgaagctttt ttctcagaat
 541 gaagtgtacc ctaactagcc gaggaagaac tatgaacata aagtctgcaa catggaaggt
 601 attgcactgc acaggccaca ttcacgtata tgataccaac agtaaccaac ctcagtgtgg
 661 gtataagaaa ccacctatga cctgcttggt gctgatttgt gaacccattc ctcacccatc
 721 aaatattgaa attootttag atagcaagac tttootcagt cgacacagco tggatatgaa
 781 attttcttat tgtgatgaaa gaattaccga attgatggga tatgagccag aagaactttt
 841 aggeogetea atttatgaat attateatge tttggaetet gateatetga ecaaaactea
 901 tcatgatatg tttactaaag gacaagtcac cacaggacag tacaggatgc ttgccaaaag
 961 aggtggatat gtctgggttg aaactcaagc aactgtcata tataacacca agaattctca
1021 accacagige attigitating transfer transfer attation accacation
1081 tttctccctt caacaaacag aatgtgtcct taaaccggtt gaatcttcag atatgaaaat
1141 gactcagcta ttcaccaaag ttgaatcaga agatacaagt agcctctttg acaaacttaa
1201 gaaggaacct gatgetttaa etttgetgge cecageeget ggagacacaa teatatettt
1261 agattttggc agcaacgaca cagaaactga tgaccagcaa cttgaggaag taccattata
1321 taatgatgta atgctcccct cacccaacga aaaattacag aatataaatt tggcaatgtc
1381 tocattacco accgetgaaa egecaaagee acttegaagt agtgetgaee etgeaeteaa
1441 tcaagaagtt gcattaaaat tagaaccaaa tccagagtca ctggaacttt cttttaccat
1501 gccccagatt caggatcaga cacctagtcc ttccgatgga agcactagac aaagttcacc
1561 tgagcctaat agtcccagtg aatattgttt ttatgtggat agtgatatgg tcaatgaatt
1621 caagttggaa ttggtagaaa aactttttgc tgaagacaca gaagcaaaga acccattttc
1681 tactcaggac acagatttag acttggagat gttagctccc tatatcccaa tggatgatga
1741 cttccagtta cgttccttcg atcagttgtc accattagaa agcagttccg caagccctga
1801 aagcgcaagt cctcaaagca cagttacagt attccagcag actcaaatac aagaacctac
1861 tgctaatgcc accactacca ctgccaccac tgatgaatta aaaacagtga caaaagaccg
1921 tatggaagac attaaaatat tgattgcatc tccatctcct acccacatac ataaagaaac
1981 tactagtgcc acatcatcac catatagaga tactcaaagt cggacagcct caccaaacag
2041 agcaggaaaa ggagtcatag aacagacaga aaaatctcat ccaagaagcc ctaacgtgtt
2101 atctgtcgct ttgagtcaaa gaactacagt tcctgaggaa gaactaaatc caaagatact
2161 agetttgeag aatgeteaga gaaagegaaa aatggaacat gatggtteae ttttteaage
2221 agraggaatt ggaacattat tacagcagcc agacgatcat gcagctacta catcacttic
2281 ttggaaacgt gtaaaaggat gcaaatctag tgaacagaat ggaatggagc aaaagacaat
2341 tattttaata coctotgatt tagcatgtag actgotgggg caatcaatgg atgaaagtgg
2401 attaccacag ctgaccagtt atgattgtga agttaatgct cctatacaag gcagcagaaa
2461 cctactgcag ggtgaagaat tactcagagc tttggatcaa gttaactgag cttttctta
2521 atttcattcc tttttttgga cactggtggc tcactaccta aagcagtcta tttatatttt
2581 ctacatctaa ttttagaagc ctggctacaa tactgcacaa acttggttag ttcaatttt
2641 gatccccttt ctacttaatt tacattaatg ctcttttta gtatgttctt taatgctgga
2701 teacagaeag eteatitiet eagitititig glatitaaae caltigeatig eaglageate
2761 attttaaaaa atgcaccttt ttatttattt atttttggct agggagttta tccctttttc
2821 gaattatttt taagaagatg ccaatataat ttttgtaaga aggcagtaac ctttcatcat
2881 gatcataggo agttgaaaaa tttttacaco tttttttca cattttacat aaataataat
2941 getttgecag cagtaegtgg tagecacaat tgeacaatat attttettaa aaaataecag
3001 cagttactca tggaatatat tctgcgttta taaaactagt ttttaagaag aaatttttt
3061 tggcctatga aattgttaaa cctggaacat gacattgtta atcatataat aatgattctt
3121 aaatgctgta tggtttatta tttaaatggg taaagccatt tacataatat agaaagatat
3181 gcatatatet agaaggtatg tggcatttat ttggataaaa tteteaatte agagaaatea
3241 tetgatgttt etatagteae tttgecaget caaaagaaaa caataceeta tgtagttgtg
3301 gaagtttatg ctaatattgt gtaactgata ttaaacctaa atgttctgcc taccctgttg
3361 gtataaagat attttgagca gactgtaaac aagaaaaaaa aaatcatgca ttcttagcaa
3421 aattgoctag tatgitaatt tgotcaaaat acaatgittg attitatgoa cittgioget
3481 attaacatcc tttttttcat gtagatttca ataattgagt aattttagaa gcattattt
3541 aggaatatat agttgtcaca gtaaatatct tgttttttct atgtacattg tacaaatttt
3601 teatteettt tgetettigt ggttggatet aacactaaet gtattgtttt gttacateaa
3661 ataaacatct tctgtgga
```

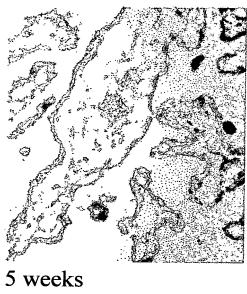
Inventor: CANIGGIA ET AL.
Docket No.: 11757.38USD1
Title: METHODS TO DIAGNOSE A REQUIRED REGULATION OF TROPHOBLAST INVASION
Serial No.: 10/028,158
Sheet 5 of 21

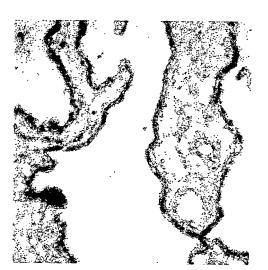
FIG. 3A



Inventor: CANIGGIA ET AL.
Docket No.: 11757.38USD1
Title: METHODS TO DIAGNOSE A REQUIRED REGULATION OF TROPHOBLAST INVASION
Serial No.: 10/028,158
Sheet 6 of 21

# FIG. 3B

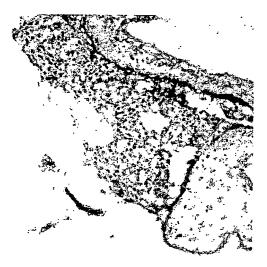




12 weeks



8 weeks



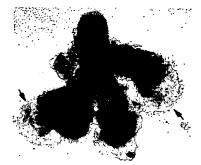
8 weeks (control)

Inventor: CANIGGIA ET AL.
Docket No.: 11757.38USD1
Title: METHODS TO DIAGNOSE A REQUIRED REGULATION OF TROPHOBLAST INVASION
Serial No.: 10/028,158
Sheet 7 of 21

FIG. 4A



AS-ß3

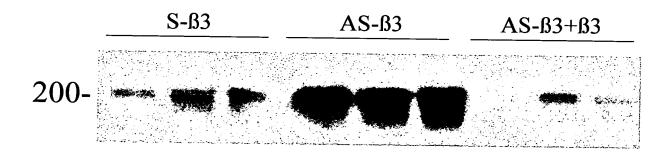


AS-\(\beta 3 + \beta 3\)



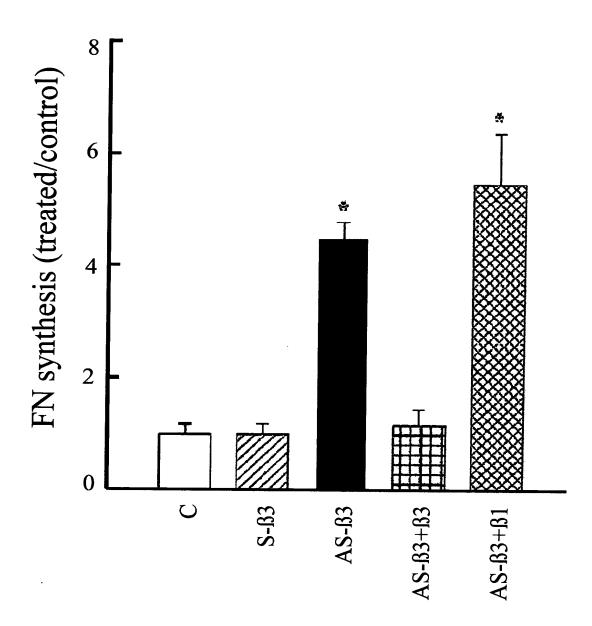
Inventor: CANIGGIA ET AL.
Docket No.: 11757.38USD1
Title: METHODS TO DIAGNOSE A REQUIRED REGULATION OF TROPHOBLAST INVASION
Serial No.: 10/028,158
Sheet 8 of 21

FIG. 4B



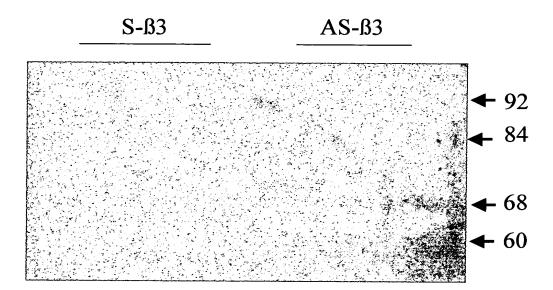
Inventor: CANIGGIA ET AL.
Docket No.: 11757.38USD1
Title: METHODS TO DIAGNOSE A REQUIRED REGULATION OF TROPHOBLAST
INVASION
Serial No.: 10/028,158
Sheet 9 of 21

FIG. 4C



Inventor: CANIGGIA ET AL.
Docket No.: 11757.38USD1
Title: METHODS TO DIAGNOSE A REQUIRED REGULATION OF TROPHOBLAST INVASION
Serial No.: 10/028,158
Sheet 10 of 21

FIG. 4D



Inventor: CANIGGIA ET AL.
Docket No.: 11757.38USD1
Title: METHODS TO DIAGNOSE A REQUIRED REGULATION OF TROPHOBLAST INVASION

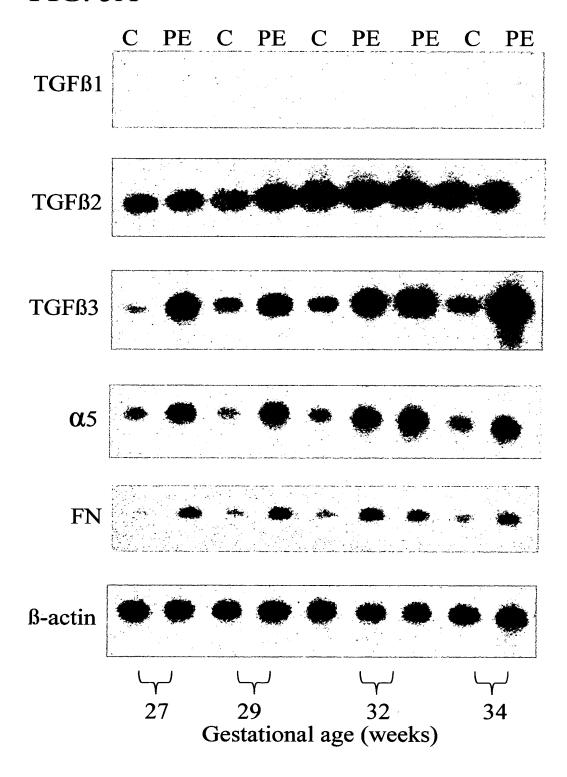
Serial No.: 10/028,158 Sheet 11 of 21

FIG. 4E



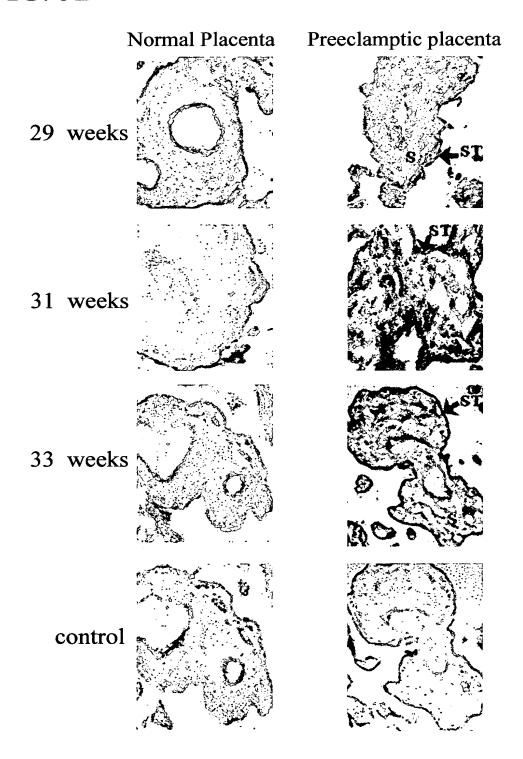
Inventor: CANIGGIA ET AL.
Docket No.: 11757.38USD1
Title: METHODS TO DIAGNOSE A REQUIRED REGULATION OF TROPHOBLAST
INVASION
Serial No.: 10/028,158
Sheet 12 of 21

### FIG. 5A



Inventor: CANIGGIA ET AL.
Docket No.: 11757.38USD1
Title: METHODS TO DIAGNOSE A REQUIRED REGULATION OF TROPHOBLAST INVASION
Serial No.: 10/028,158
Sheet 13 of 21

## FIG. 5B

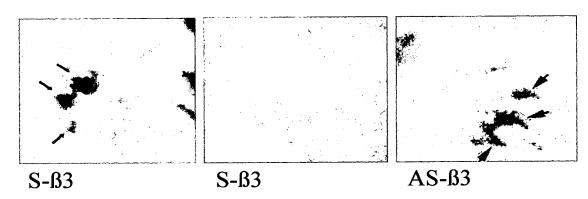


Inventor: CANIGGIA ET AL.
Docket No.: 11757.38USD1
Title: METHODS TO DIAGNOSE A REQUIRED REGULATION OF TROPHOBLAST INVASION
Serial No.: 10/028,158
Sheet 14 of 21

## FIG. 6A

#### Normal Placenta

#### Preeclamptic placenta

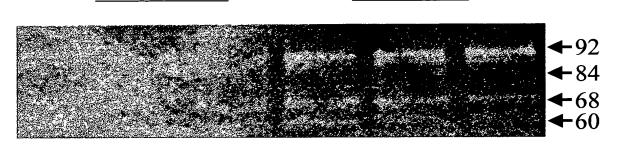


Inventor: CANIGGIA ET AL.
Docket No.: 11757.38USD1
Title: METHODS TO DIAGNOSE A REQUIRED REGULATION OF TROPHOBLAST INVASION
Serial No.: 10/028,158
Sheet 15 of 21

FIG. 6B

S-ß3

AS-B3



Inventor: CANIGGIA ET AL.
Docket No.: 11757.38USD1
Title: METHODS TO DIAGNOSE A REQUIRED REGULATION OF TROPHOBLAST INVASION
Serial No.: 10/028,158
Sheet 16 of 21

FIG. 6C

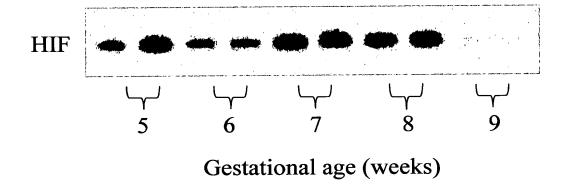
S-ß3

AS-ß3



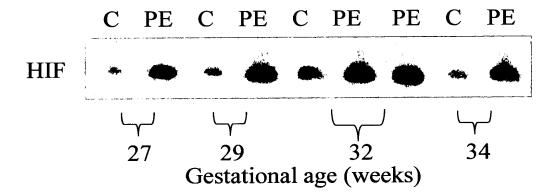
Inventor: CANIGGIA ET AL.
Docket No.: 11757.38USD1
Title: METHODS TO DIAGNOSE A REQUIRED REGULATION OF TROPHOBLAST
INVASION
Serial No.: 10/028,158
Sheet 17 of 21

FIG. 7A



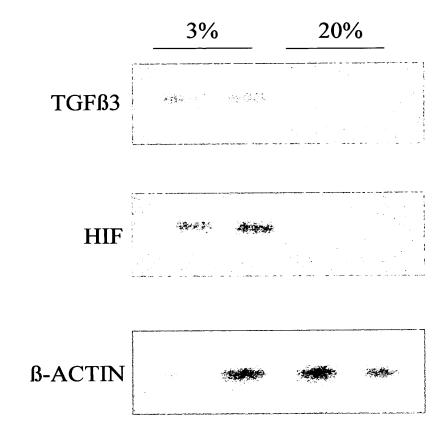
Inventor: CANIGGIA ET AL.
Docket No.: 11757.38USD1
Title: METHODS TO DIAGNOSE A REQUIRED REGULATION OF TROPHOBLAST
INVASION
Serial No.: 10/028,158
Sheet 18 of 21

## FIG. 7B



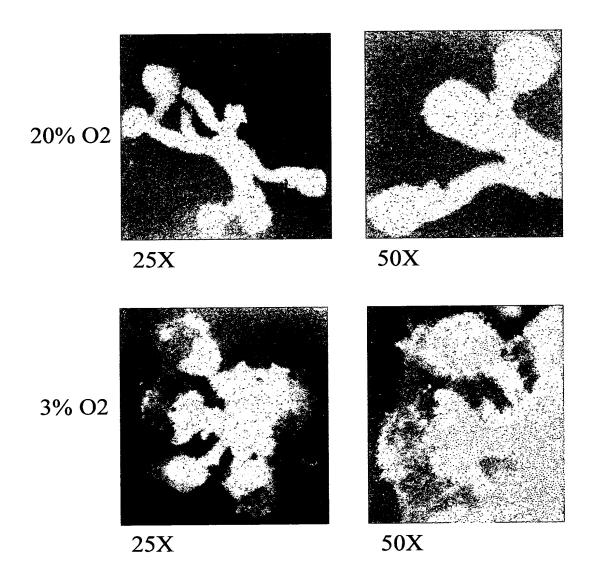
Inventor: CANIGGIA ET AL.
Docket No.: 11757.38USD1
Title: METHODS TO DIAGNOSE A REQUIRED REGULATION OF TROPHOBLAST INVASION
Serial No.: 10/028,158
Sheet 19 of 21

## FIG. 8



Inventor: CANIGGIA ET AL.
Docket No.: 11757.38USD1
Title: METHODS TO DIAGNOSE A REQUIRED REGULATION OF TROPHOBLAST
INVASION
Serial No.: 10/028,158
Sheet 20 of 21

FIG. 9



Inventor: CANIGGIA ET AL.
Docket No.: 11757.38USD1
Title: METHODS TO DIAGNOSE A REQUIRED REGULATION OF TROPHOBLAST INVASION
Serial No.: 10/028,158
Sheet 21 of 21

FIG. 10

